CLAIMS

1. A compound with the following formula (I):

in which:

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$$A = \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \text{or} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_1 \\ R_2 \\ R_2 \\ R_2 \end{array} \qquad \begin{array}{c} R_2 \\ R_2$$

- R₁, R₂ = H, -CH₃, -CH(CH₃)₂, -OCH₃, -Cl, -CF₃, -OCF₃, -SCH₃;
- R_3 = an amino acid radical hydrolysable by a carboxypeptidase A;
- $R_4 = a$ basic amino acid radical.
- 2. A compound according to claim 1 with the following formula (I):

in which:

- R₁, R₂ = H, -CH₃, -CH(CH₃)₂, -OCH₃, -Cl, -CF₃, -OCF₃, -SCH₃;
- R₃ = a hydrophobic amino acid radical;
- R₄ = an arginine or lysine radical.
- 5 3. A compound according to claim 1 or claim 2, characterized in that $R_1 = H$ and $R_2 = -S-CH_3$.
 - 4. A compound according to claim 1 or claim 3, characterized in that R₃ is selected from the following amino acids:
 - tyrosine:
- phenylalanine;
 - alanine;
 - valine;
 - leucine;
 - isoleucine;
 - phenylglycine.

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- 5. A compound according to claim 1 or claim 3, characterized in that R_3 represents phenylalanine.
- 6. A compound according to claim 1 or claim 3, characterized in that R₃ represents phenylalanine or tyrosine and R₄ represents arginine or lysine.
- 20 7. A compound according to claim 1 or claim 3, characterized in that R₃ represents tyrosine.
 - 8. A compound according to claim 1, characterized in that R₁ is selected from: -H and -CH₃, and R₂ is selected from CH₃, O-CH₃ and -S-CH₃.
 - 9. A compound according to any one of claims 1 to 8, characterized in that A is:

R₁

10. A compound according to claim 1 with formula (I), in which:

said compound being selected from the group constituted by the following compounds in which:

$$R_{1} = -CH_{3} \ _{GF} \qquad R_{2} = -CH_{3} \ _{GF} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -CH_{3} \ _{GF} \qquad R_{2} = -CH_{3} \ _{GF} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -CH_{3} \ _{GF} \qquad R_{2} = -CH_{3} \ _{GF} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -C - CH_{3} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -CH_{3} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -CH_{3} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -CH_{3} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -CH_{3} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -CH_{3} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -CH_{3} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -CH_{3} \qquad R_{3} = -CH_{2} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{2}$$

$$R_{1} = -H \qquad R_{2} = -CH_{3} \qquad R_{3} = -CH_{2} \qquad NH - C NH_{2}$$

$$R_{2} = -(CH_{2})_{3} - NH - C NH_{2} \qquad NH_{2} \qquad NH_{3} = -CH_{3} \qquad R_{4} = -(CH_{2})_{3} - NH - C NH_{3} \qquad NH_{4} \qquad NH_{4} = -CH_{2} \qquad NH_{4} \qquad NH_{4} \qquad NH_{4} \qquad NH_{4} \qquad NH_{4} = -CH_{4} \qquad NH_{4} \qquad N$$

* the numbers in brackets determining the position of the methyl groups on the phenyl radical.

25 11. A compound according to claim 1, characterized in that it is 4-MTPAFYR (4-methylthiophenylazoformyltyrosine arginine).

- 12. A method for assaying the activity of a carboxypeptidase N or a carboxypeptidase U in a biological sample, in which:
 - said sample is brought into contact with a compound with formula (I) according
 to any one of claims 1 to 11, and with a carboxypeptidase A, under conditions that
 allow hydrolysis of the sample; and
 - the reduction in coloration of the sample containing the substrate with formula (I) and carboxypeptidase A is measured, resulting from double hydrolysis of the substrate with formula (I) by the CPN or CPU of the sample and by CPA.
- 13. A method according to claim 12, characterized in that $R_1 = H$ and $R_2 = -S-CH_3$.
- 10 14. A method according to claim 12 or claim 13, characterized in that R₄ is an arginine or lysine radical.
 - 15. A method according to claim 12 or claim 13, characterized in that the substrate is a compound with formula (I) in which R₃ is selected from the following amino acid radicals:

• tyrosine:

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- phenylalanine;
- alanine;
- valine;
- leucine;
- isoleucine;
- phenylglycine.
- 16. A method according to one of claims 12 to 15, characterized in that R₃ is tyrosine.
- 17. A method according to claims 12 to 15, characterized in that the substrate is a compound with formula (I), in which R₃ represents phenylalanine.
- 25 18. A method according to claims 12 to 15, characterized in that the substrate is a compound with formula (I) in which R₃ represents phenylalanine and R₄ represents arginine or lysine.

- 19. A method according to any one of claims 21 to 18, characterized in that the substrate is a compound with formula (I) in which R₁ is selected from -H and -CH₃, and R₂ is selected from CH₃, O-CH₃ and -S-CH₃.
- 20. A method according to claim 12, characterized in that the substrate is a compound with formula (I) in which:

in which:

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- R_1 , $R_2 = H$, $-CH_3$, $-CH(CH_3)_2$, $-OCH_3$, -Cl, $-CF_3$, $-OCF_3$, $-SCH_3$;
- R_3 = an amino acid radical hydrolysable by a carboxypeptidase A;
- R₄ = a basic amino acid radical.
- 21. A method according to claim 21, characterized in that the substrate is a compound with formula (I) in which:

said compound being selected from the group constituted by the following compounds:

$$R_{4} = -(CH_{2})_{3} - NH - CNH_{2}$$

$$R_{1} = -H$$

$$R_{2} = -CH_{3}$$

$$R_{3} = -CH_{2} - CH_{2}$$

$$R_{4} = -(CH_{2})_{3} - NH - CNH_{2}$$

$$R_{4} = -(CH_{2})_{3} - NH - CNH_{2}$$

$$R_{4} = -(CH_{2})_{4} - NH_{2}$$

$$R_{1} = -H$$

$$R_{2} = -CH_{3}$$

$$R_{3} = -CH_{2} - CH_{2} - NH_{2}$$

$$R_{4} = -(CH_{2})_{4} - NH_{2}$$

$$R_{1} = -H$$

$$R_{2} = -CH_{3}$$

$$R_{3} = -CH_{2} - CH_{2} - NH_{2}$$

$$R_{4} = -(CH_{2})_{3} - NH_{3} - CH_{2} - NH_{4}$$

$$R_{4} = -(CH_{2})_{3} - NH_{3} - CH_{4} - NH_{4}$$

$$R_{4} = -(CH_{2})_{3} - NH_{3} - CH_{4} - NH_{4}$$

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$$R_{4} = -(CH_{2})_{3} - NH_{4} - CH_{4} - CH_{4}$$

$$R_{4} = -(CH_{2})_{3} - NH_{4} - CH_{4}$$

$$R_{4} = -(CH_{2})_{$$

$$R_1 = -H$$
 $R_2 = -SCH_3$ $R_3 = -CH_2$ $R_4 = -(CH_2)_3 - NH - C$ NH_2 $R_1 = -H$ $R_2 = -SCH_3$ $R_3 = -CH_2$ $-OH$ $R_4 = -(CH_2)_3 - NH - C$

- *the numbers in brackets determining the position of the methyl groups on the phenyl radical.
 - 22. A method according to any one of claims 12, 13, 15, 20 or 21, in which the compound with formula (I) is 4-MTPAFYR (4-methylthiophenylazoformyltyrosine arginine).
 - 23. A method according to any one of claims 12 to 22, characterized in that the optical density of the mixture is measured without adding CPA, then after adding CPA.
 - 24. A method according to any one of claims 12 to 23, characterized in that the measured decrease in coloration is compared with values on a calibration curve.
 - 25. A method according to any one of claims 12 to 24, characterized in that the sample is a blood sample.
- 15 26. A method according to claim 25, characterized in that the sample is plasma.

- 27. A method according to any one of claims 12 to 26, characterized in that the CPA is pancreatic CPA.
- A method according to any one of claims 12 to 27, characterized in that the test sample is brought into the presence of an activator buffer for the time necessary to obtain activation of the carboxypeptidase U the activity of which is to be measured, then into the presence of a protease serine inhibitor.
- A method according to claim 28, characterized in that the substrate with formula (I) is added at the same time as the activator buffer, or simultaneously or immediately after the serine protease inhibitor.
- 25 30. A method according to claim 28, characterized in that activation is carried out using the thrombin/thrombomodulin complex route.

- A method for assaying the activity of the constitutional CPN or CPU of a sample and that of the activatable CPN or CPU of the same sample, characterized in that the hydrolysis activity of the sample on a sample with formula (I) is compared after bringing the sample into the presence of an activator buffer, if necessary for the time necessary to obtain activation of the carboxypeptidase U the activity of which is to be measured, then into the presence of a protease serine inhibitor, the observed hydrolysis activity being compared with the hydrolysis activity of the sample on a substrate with formula (I) in the absence of an activator buffer in accordance with claim 21.
- 32. A method according to any one of claims 21 to 28, characterized in that the carboxypeptidase is a CPU.
 - 33. A method according to claim 32, characterized in that the CPU is TAFI.

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- 34. A method according to any one of claims 28 to 33, characterized in that the sample is treated in the presence and in the absence of a specific TAFI inhibitor.
- 35. A method according to any one of claims 28 to 34, characterized in that the specific TAFI inhibitor is CPI.
 - 36. A method for assaying activated TAFI in a blood sample, comprising the following steps:
 - a) bringing a first aliquot of the sample into contact with a specific TAFI inhibitor and treating it using the method defined in claim 28;
 - b) treating a second aliquot of the sample using the method of claim 28, in the absence of specific TAFI inhibitor;
 - c) measuring the Δ OD between the first and second aliquot, representative of the activity of the activated TAFI in the sample.
- A method according to claim 36 for differentiating between the activity of constitutional TAFI and that of activatable TAFI in the same sample, characterized in that the hydrolysis activity of a third aliquot of the sample is measured on a substrate with formula (I) in the absence of a buffer activator.

- 38. Use of a compound with formula (I) according to any one of claims 1 to 11, to assay the enzymatic activity of a carboxypeptidase N or U in a sample.
- 39. Use according to claim 38, characterized in that the carboxypeptidase is TAFI.
- 40. A kit for assaying the activity of a CPN or a CPU in a sample comprising a chromogenic substrate constituted by a compound according to any one of claims 1 to 11.
- 41. A kit for assaying the activity of TAFI in a biological sample, comprising:
 - a TAFI activator buffer;
 - carboxypeptidase A;
 - a substrate with formula (I) according to any one of claims 1 to 11;
- a TAFI inhibitor.